A RAPID LABORATORY METHOD FOR TESTING KEROSENE-BASE INSECTICIDES AGAINST HOUSE FLIES

By F. L. Campbell and W. N. Sullivan,
Division of Insect Physiology and Toxicology,
Bureau of Entomology, U. S. Department of Agriculture

Liquid household insecticides are usually tested in cubical chambers 6 feet on a side by the method described by Peet and Grady. 1/ This method was intended to give reproducible results and at the same time to simulate conditions under which fly sprays are applied in practice. However, it has not made practical tests unnecessary and has come to be used chiefly for controlling the quality of commercial fly sprays, for determining the relative value of different brands of household insecticides, and for developing new insecticides. For these purposes the Peet-Grady method might well be replaced by one that is more rapid, provided it gives equally dependable results. It is the purpose of this article to describe such a method in sufficient detail to permit anyone interested to try it. This method, which is a modification of one recently described by the writers, 2/ is very simple but requires two people to operate it. Later the writers will describe a further modification of this method that costs more for equipment but can be operated by one man.

Equipment Necessary for Making 36 Tests per Day on 3,600 Flies

(Equipment and supplies for rearing are not included)

Cold room for chilling flies at 32° F. An ordinary electric household refrigerator is not satisfactory for this purpose.

Two heavy-walled ointment jars or any well-insulated container for holding chilled flies in reserve for counting.

Thirty-six Petri dishes (bottoms only) 143 mm outside diameter and 15 mm deep.

Wire screen (14 mesh) for covering Petri dishes.

Celluloid bands, 1/2 inch wide, for holding the wire screen snugly on the dishes. The bands are made by cutting out strips of celluloid (15/1,000 inch thick) in proper lengths and sealing the ends together by an acetone solution of celluloid.

1/ Peet, C. H., and Grady, A. C. 1928. Studies in Insecticidal Activity. I. Testing Insecticides Against Flies. Jour. Econ. Ent. 21: 612-617.

2/ Campbell, F. L., Sullivan, W. N., and Jones, H. A. 1934. Kerosene Extracts of Derris Root as House Fly Sprays. Part I. Method and Results of Laboratory Tests of Extracts of Derris and of Cube Roots. Soap, vol. 10, no. 3, pp. 81, 82, 83, 85, 87, 103, 105, 107.

Circles of 15 cm filter paper of the cheapest grade for covering the bottoms of the dishes.

Six glass cylinders, 17½ inches high and 8¼ inches inside diameter.

Six window glass plates, 9½ by 9½ inches, for covering cylinders.

Six window glass plates, 10 by 15 inches, on which cylinders stand.

One bell jar with open top, tubulation near bottom and wide, well ground flange, 200 mm high and 200 mm diam. Be sure that ground flange will fit on cylinders.

One paint spray gun to be mounted rigidly on top of bell jar with nozzle of gun in hole of bell jar. Two wooden blocks may be clamped together by nuts and bolts about the neck of the bell jar, and the gun may be attached vertically to the blocks by right-angle iron braces.

One right-angle liquid inlet tube for spray gun and cork on tube to hold the glass tube containing the liquid to be sprayed. The cork should be notched or drilled to admit air.

Not less than six 15 c c graduated centrifuge tubes to hold the liquid to be sprayed.

Six feet of hose, reducing valve, and ouncer gauge attached to compressed air line.

One table, 6 feet long and 2 feet wide, on which cylinders are arranged in line.

One small table for transferring flies from dishes to cages.

Thirty-six observation cages. The writers use a 9½ inch cubical cage having a sliding glass front, sheet rubber back (made from discarded inner tubes of automobile tires), and top and sides covered with wire screen. An arm hole (1¾ inches diameter) is cut in the center of the rubber back and is closed with the lid of a screw-top jar.

Two warm constant-temperature rooms, one in which treatments are made and the other in which flies are reared and observed after treatment. Cylinders could be set up within a Peet-Grady chamber if no other room were available, but it would be desirable to have a larger room with higher ceiling for the treatments.

Operation of Method

No changes have been made in the previously described 3/ preparations

3/ See footnote 2.

for treatments except that flies are now being maintained in the ordinary way without special provision for artificial illumination and 100 flies instead of 50 are being used in each Petri dish.

At the beginning of a series of six tests the bell jar is placed on the cylinder at the left end of the line. The two operators will be called A and B. The centrifuge tube containing 5 c c of the liquid to be sprayed is attached by A to the cork stopper on the inlet tube of the spray gun so that the end of the inlet tube coincides with the 1/2 c c mark in the conical end of the centrifuge tube. The valve of the spray gun is opened by A for 5 seconds (pressure while spraying 10 pounds per square inch), all the liquid within reach of the tube being sucked up within that period and about 4 c c being sprayed into the bell jar and cylinder, filling the chamber with a dense mist. Immediately after spraying, A lifts the bell jar slightly while B slips a glass plate over the cylinder. A then places the bell jar on the plate over the second cylinder and tilts the first cylinder back as B slips a dish of flies under it. A drops the cylinder back over the dish and centers the dish by shifting the position of the cylinder if necessary, while B starts a stop watch. A removes the centrifuge tube, B picks up the bell jar and opens the valve, and A holds a beaker of acetone to the inlet tube of the spray gun for a few seconds, B spraying and wiping out the bell jar with cheesecloth to clean it. A removes the plate from the top of the second cylinder and B places the bell jar on the second cylinder, completing the cycle. Then, as before, A adjusts the next centrifuge tube on the spray gun, sprays, and so on, and B records the time on the stop watch at which the second dish of flies was exposed. Thus the six treatments are made in order and are usually completed in 5 minutes. During the next 5 minutes the observation cages are made ready to receive the flies and another lot of six dishes of flies is brought into the warm room for the next series of tests. At the end of 10 minutes after the first treatment the dish of flies under the first cylinder is withdrawn and the flies are transferred to a cage, as previously described. 4/ The other dishes are then withdrawn in order at the end of each 10-minute exposure period and the flies transferred to the cages, which are then placed in an adjacent constant-temperature room. The mist still remaining in the cylinders is blown out and all glassware is wiped with cheesecloth. Meanwhile the centrifuge tubes are refilled, completing preparations for the next series of tests, which are then made as just described. Six series of tests, 36 tests in all, can be made in less than 2 hours.

Discussion

The particulars of the foregoing method, i.e., volume of liquid sprayed, pressure, period of exposure, etc., do not necessarily represent the most efficient combination. They were chosen because they gave very low mortality in the kerosene checks and a kill with pyrethrum sprays similar to those obtained in the Peet-Grady chamber. The writers are not stressing particular details here so much as the general principle of progressive treatments in a row of small chambers.

4/ See footnote 2.

The principal advantage of the present method over the Peet-Grady method is in speed. To make 15 tests a day in a single Peet-Grady chamber is considered fast work. The number that can be made by the present method is limited only by facilities for rearing and caging flies. The writers know that 36 tests can be made easily in a day, including preparations and observations, all but the counting of chilled flies and the treatments being done by one man.

The preparation of the flies for testing and the relatively short period of the treatments in the writers' method have at least a theoretical advantage over the practice in the Peet-Grady method. In the writers' method all flies for the day's work are drawn at random from a single large lot of mixed chilled flies; whereas in the Peet-Grady method the flies may be taken from different cages over a relatively long period of time. Variation in results of tests of a single liquid should therefore be less in the writers' method, though this has not yet been proved.

In the writers' method all flies treated are kept for observation, whereas in the Peet-Grady method only those knocked down are retained, to the disadvantage of substances that do not take effect rapidly.

Neither the writers' method nor any other method should be used for evaluating fly sprays without including a standard insecticide in each series of tests. With the same liquid one might kill from 25 to 75 percent of the flies, depending on their susceptibility, which cannot be controlled and standardized. Therefore, to say that a certain fly spray killed 60 percent of the flies by a certain method means nothing. The best method of using a standard insecticide remains to be worked out. The writers are inclined to believe that comparisons should be based on a 50 percent kill.